## REMARKS

This Reply is in response to the Office Action dated February 4, 2003. This Reply is filed along with a petition for a one month extension of time along with an authorization to charge the required statutory fee.

Claims 1-12 were pending at the time of the Office Action. In the Office Action, claims 1-12 were all rejected. In this Reply, Claims 1-12 have been amended and Claims 13-19 have been added. No new matter has been added. The amended claims are shown in a section entitled "Marked-Up Version to Show Changes Made" using standard underlining and bracketing format to highlight the changes made.

Claim 5 was rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which Applicant regards as the invention based on the recital of "etc." In response, Applicant has amended claim 5 to remove "etc.". Accordingly, the 5 U.S.C. 112, second paragraph of claim 5 is now overcome.

Now turning to rejections based on art, claims 1 and 5-12 were rejected under 35 U.S.C. 102(b) as anticipated by or, in the alternative, under 35 U.S.C. 103(a) as obvious over Chong (Anal. Chem., 69:3889-3898 (1997). Claims 2 and 3 were rejected under 35 U.S.C. 103(a) as being unpatentable over Chong in view of either Kataoka (Anal. Chem., 71:4237-4255 (October 1, 1999)) (Kataoka I) or (Chromatographia, 50(9/10):532-538 (November, 1999)) (Kataoka II) and either Wang (Anal. Chem., 69:4566-4576 (1997)), or Malik (Advanced Sol-gel Column Technology for Condensed-phase Microseparations, pg. 54 (1997).

Claim 4 was rejected under 35 USC 103(a) as being unpatentable over Chong in view of either Kataoka I, Kataoka II, and either Wang or Malik as applied to Claims 2 and 3 above, and further in view of either Malik or Nakanishi (U. S. Patent No. 5,624,875). Claim 9 was rejected under 35 U.S.C. 103(a) as being unpatentable over Chong in view of Kataoka I.

Claims 1-12 were rejected under 35 U.S.C. 103(a) as being unpatentable over Kataoka I, in view of Chong and either Wang or Malik. Claim 4 was rejected under 35 USC 103(a) as being unpatentable over Kataoka I in view of Chong and either Wang or Malik as applied to Claims 1-12 above, and further in view of either Malik or Nakanishi.

Before reviewing the cited art, Applicant will first review the claimed invention as recited in amended claim 1. Amended claim 1 recites a method of preconcentrating trace analytes and includes the steps of providing a hollow capillary having at least one sol-gel extraction medium

within the hollow capillary. The sol-gel extraction medium is chemically bound to inner walls of the hollow capillary to form a sol-gel extraction medium-loaded capillary (e.g. page 32, lines 4-11). The loaded capillary is exposed to a sample containing at least one target analyte, wherein the target analyte becomes disposed inside the hollow capillary. Amended claim 4 recites the sol-gel extraction medium comprises a porous sol gel monolithic bed.

The invention expands the scope of in-tube solid-phase microextraction (in-tube SPME) as well as SPME in general by introducing sol-gel coatings and porous monolithic beds as extraction medium in in-tube SPME. None of the cited art discloses the claimed sol-gel coatings for application to in-tube SPME. The invention also solves a significant and often limiting problem in in-tube SPME, namely providing operational stability (solvent stability and thermal stability) to the extraction medium, which can be a coating or porous monolithic bed, by *chemically binding* the sol-gel based extraction medium to the inner walls of the capillary. Due to chemical bonding, sol-gel coatings have significant advantages over their conventional counterparts in terms of both operational lifetime and extraction reproducibility.

The claimed invention also allows the creation of highly stable coatings that are a few orders of magnitude greater in thickness than previous work. The stability is achieved through chemical bonding of the coating to the capillary inner walls. Sol-gel coatings possess higher surface area compared with their conventional counterparts. The higher coating thickness and enhanced surface area allows sol-gel coatings to provide significantly higher sample capacity and extraction sensitivity compared with their conventional counterparts. For example, page 42, lines 22-24 discloses that parts per trillion (ppt) and parts per quadrillion (ppq) level detection sensitivities are achieved by the invention for both polar and non-polar analytes.

Since the claims have been significantly amended, Applicant will review each of the cited references and discuss distinctions of the claimed invention (extraction method, extraction device and method of forming an extraction device) as appropriate as compared to the cited art.

Chong is a publication in which the author of the present patent application is the lead (corresponding) author. The fiber-based microextraction format described in Chong is quite different from the capillary microextraction method and device claimed in the present application. For example, in the fiber-based format of Chong the sol-gel coating is applied to the *outer surface of a solid rod* whereas in the claimed capillary microextraction format of the

present invention, the sol-gel extraction medium is chemically bound *within a hollow capillary* either in the form of a sol-gel surface coating or a porous sol-gel monolithic bed.

The Kataoka I and II references each disclose coated capillaries for performing in-tube solid-phase microextraction, as does the claimed invention. However, the devices and methods disclosed by both Kataoka references significantly differ from the claimed invention. The Kataoka references use gas chromatography (GC) capillary columns with a polymeric stationary phase coating for extraction. The Kataoka references do not disclose or suggest use of a sol-gel media for the stationary phase coating. Moreover, the stationary polymeric phase coatings inside Kataoka's capillaries are *physically held*. In contrast, the claimed invention recites a hollow tube having *chemically bonded sol-gel medium* therein. In addition, both Kataoka references disclose only surface coatings as extraction media. Thus, use of the monolithic bed recited in method claim 4 and related device claim 13 adds an additional patentable feature to the claimed invention.

There are several drawbacks in using the coated GC capillaries disclosed by Kataoka. First, as noted above the stationary phase coatings in such capillaries are physically held, *not* secured though chemical bonding to the inner wall of the capillary as claimed by Applicant. As a result, the physically held stationary phase coatings are quickly stripped off the capillary during the course of normal operations. This, in turn, shortens the useful lifetime of the extraction capillary and causes reproducibility problems. Second, the coatings inside the GC columns are very thin, the most frequently used coating thickness is about 0.25 μm. Such thin coatings limit the sample capacity (and hence extraction sensitivity) of conventional in-tube SPME. It is extremely difficult to create a stable, thick stationary phase coating on the inner surface of the capillary using conventional coating technologies.

Malik discloses the use of very small diameter sol-gel columns (25-μm and 50-μm internal diameter) for capillary electrophoretic separations, not for in-tube SPME or for sample preconcentration in general. Microseparation techniques are far different from microextraction techniques as are the devices disclosed by Malik for capillary electrophoretic separations. It is well known in the art that capillary electrophoresis techniques require that the separation column be of a sufficiently small internal diameter to avoid excessive Joule heating. For extraction, however, such small diameter capillaries are not suitable since the sample capacity of such small

diameter capillaries will be exceedingly low which results in severely diminished sample capacity and extraction sensitivity.

No mention is made in Malik as to whether the disclosed capillary electrophoresis columns can be used for extraction. However, Malik teaches away from use of the disclosed structure therein for extraction. As noted above, exceedingly small diameter of the disclosed capillary electrophoresis columns required to minimize Joule heating in the column makes the sample capacity of such capillaries impractical for use as a sample extraction and/or preconcentration device.

In contrast, Applicant discloses a 250-μm internal diameter capillary in FIGs. 11 and 15 and accompany disclosure which is now claimed in device claim 15 (added). Device claim 12 (added) recites the device providing at least parts per trillion (ppt) level detection sensitivities. Thus, the capacity and resulting detection sensitivity of Malik's columns if used for extraction would be approximately one hundred times smaller than that of Applicant's disclosed extraction capillaries.

Wang discloses gas chromatography (GC) separation columns which include coated capillaries. The disclosed GC separation columns have a sub-micrometer sol-gel stationary phase film thickness of about 0.25 μm. Wang does not disclose or suggest using the disclosed GC columns for extraction. The thin coating makes the capacity and resulting sensitivity of Wang's GC device poor for extraction. Thus, as with Malik, Wang teaches away from use of the disclosed separation device as an extraction device.

Nakanishi discloses sol-gel monolithic columns for use in liquid chromatography separation, not for extraction. The monolithic bed disclosed in Nakanishi is not in the capillary format. It is also unlikely the sol-gel technology described in Nakanishi can be applied to a capillary format since the diameter of the separation beds described therein are 4.6 mm and 6.0 mm respectively compared with the typical diameter of the bed in the capillary format is on the order of 0.25 mm.

The method disclosed in Nakanishi for the preparation of monolithic column is also very different from the method used in the present invention. The method developed in Nakanishi discloses a number of steps: (a) preparing sol-gel silica material using a sol solution, (b) treating the prepared sol-gel silica material with ammonia solution (for seven days), (c) heating the created sol-gel material to high temperature (600 – 800 C), (d) Using the created silica material

to prepare cylindrical rods either by placing the sol solution in a cylindrical mold (6 mm) or by mechanically shaping the solid sol-gel silica (4.6 mm diameter), (e) securing the silica rod inside a tube, (e) Derivatizing the silica rods using conventional silane chemistry to chemically bind a chromatographically favorable organic ligand to the surface of the created silica rod.

The porous monolithic bed in Nakanishi cannot be secured via chemical bonding since the tube lacks functional groups needed for chemical bonding with monolithic bed. In contrast in the claimed invention, a hollow tube (e.g. fused silica) is used to prepare the monolithic column. As disclosed in the present Application, silanol groups on the inner surface of such a tube can serve as the chemical binding sites for the porous monolithic bed in the course of its in situ creation from a sol solution. Such chemical bonding provides added operational stability and prolongs the lifetime of the sol-gel monolithic capillary.

In contrast to Nakanishi, the method of forming the extraction capillaries used in the present invention is much simpler, and essentially consists of a single-step procedure. In the claimed method of forming a microextraction device recited in claim 16, a hollow capillary and at least one sol-gel extraction medium is provided. The sol-gel extraction medium is introduced within the hollow capillary. The sol-gel extraction medium chemically bonds *in situ* to inner walls of the hollow capillary to form a sol-gel extraction medium-loaded capillary, the device providing at least parts per trillion (ppt) level detection sensitivities.

Also, the method described in Nakanishi is not adapted for the preparation of sol-gel monolithic silica beds within a fused silica capillary because Nakanishi uses high temperature heating (600-800 C). Under such high temperature conditions the protective outer coating of the fused silica capillary will be damaged rendering it very fragile for practical operation.
In view of the above, amended claim 1 and its respective dependent claims are believed to be patentable claims. Notably, claim 4 which recites the sol-gel extraction medium comprising a porous sol gel monolithic bed is believed to represent additional patentable subject matter.

Claim 12 (added) recites a microextraction device comprising a hollow capillary, and at least one sol-gel extraction medium within the hollow capillary for trapping at least one target analyte, the sol-gel extraction medium chemically bound to inner walls of the hollow capillary to form an internally coated capillary. The device provides at least parts per trillion (ppt) level detection sensitivities. Claim 13 (new) recites the sol-gel extraction medium comprising a porous sol gel monolithic bed, the monolithic bed having a thickness equal to an internal

diameter of the hollow capillary. Since the extraction sensitivity is approximately proportional to the thickness of the coating material, the embodiment recited in claim 13 provides a significantly higher sensitivity as compared to a thinly coated hollow tube. Claim 15 (new) recites the hollow capillary provides an internal diameter of at least 250 μm. In view of comments made above, Applicant submits that claim 12 and its respective dependent claims are patentable over the cited art, with many of its dependent claims representing additional patentable subject matter.

Claim 16 (added) recites a method of forming a microextraction device comprising the steps of providing a hollow capillary and at least one sol-gel extraction medium, introducing the sol-gel extraction medium within the hollow capillary, and *in situ* chemically bonding the sol-gel extraction medium to inner walls of the hollow capillary to form a sol-gel extraction medium-loaded capillary. The device formed provides at least parts per trillion (ppt) level detection sensitivities. In view of the comments made above, Applicant submits that claim 16 and its respective dependent claims are patentable over the cited art.

Applicant has made every effort to present claims which distinguish over the cited art, and it is believed that all pending claims are in condition for allowance. However, Applicant requests that the Examiner contact the undersigned after review of this Reply if the Examiner determines that any clarification is necessary to permit issuance of a Notice of Allowance.

Respectfully submitted,

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Gregory A. Nelson, Reg. No. 30,577
Neil R. Jetter, Reg. No. 46,803
AKERMAN SENTERFITT
222 Lakeview Avenue; Suite 400
P.O. Box 3188
West Palm Beach, FL 33402-3188
(561) 653-5000

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MARKED-UP VERSION TO SHOW CHANGES MADE

In the Claims

1. (Amended) A method of preconcentrating trace analytes [by], comprising the steps of:

providing a hollow capillary having at least one sol-gel extraction medium within said hollow capillary, said sol-gel extraction medium chemically bound to inner walls of said hollow capillary to form a sol-gel extraction medium-loaded capillary, and

exposing said loaded capillary to a sample containing at least one target analyte, wherein said target analyte becomes disposed inside said loaded capillary

[extracting polar and/or non-polar analytes through a sol-gel extraction medium].

2. (Amended) A method according to claim 1, wherein said [extracting] exposing step [is further redefined as] comprises [feeding a] directing said sample through [a sol-gel coated inner surface of a tube] said coated capillary [and extracting the analytes from the sample with the sol-gel coating].

3. (Amended) A method according to claim [2] 1, wherein said sol-gel extraction medium comprises a sol-gel coating [feeding step is further defined as passing the sample through a capillary tube, the tube including a sol-gel coated inner surface].

4. (Amended) A method according to claim [2] 1, wherein said sol-gel extraction medium comprises [feeding step is further defined as passing the sample through] a porous sol gel monolithic bed.

5. (Amended) A method according to claim 1, wherein [the] an organic component of [the] said sol-gel is selected from the group [including] consisting of sol-gel-active forms and/or derivatives of poly(ethylene glycol), poly(methylphenylsiloxane), poly(dimethyldiphenylsiloxane), poly(dimethylsiloxane), poly(methylcyanopropylsiloxane),

octadecylsilane, octylsilane, crown ethers, cyclodextrins, calixarenes, dendrimers, poly(styrene), poly(styrene-divinylbenzene), poly(acrylate), and molecularly imprinted polymers[, etc].

6. (Amended) A method according to claim 1, further including the step of [thermally] desorbing [the] said analyte[s] from [the] said sol-gel extraction medium to provide extracted analyte.

7. (Amended) A method according to claim [1] 6, [further including the step of desorbing the analytes from the sol-gel extraction medium] wherein said desorbing step comprises thermal desorbing.

8. (Amended) A method according to claim 6, further including the step of applying [the] said extracted analyte[s] to a GC capillary column.

9. (Amended) A method according to claim [7] 6, further including the step of [applying the] directing said extracted analyte[s] to a liquid phase separation system [technique].

10. (Amended) A method according to claim 1, further including the step[s] of preconditioning [the sol-gel] sol-gel extraction medium prior to said [extracting] exposing step.

11. (Amended) A method according to claim [8] 10, wherein said preconditioning step [is further defined as] comprises [simultaneously] heating and purging an inert gas over sol-gel extraction medium [the sol-gel].

12. (Amended) A microextraction device, comprising:

[A microextraction method including the steps of

microextraction polar and non-polar analytes in a sol-gel extraction medium;

desorbing the analytes from the sol-gel and analyzing the extracted, desorbed analytes]

a hollow capillary, and

at least one sol-gel extraction medium within said hollow capillary for trapping at least one target analyte, said sol-gel extraction medium chemically bound to inner walls of said hollow capillary to form a sol-gel extraction medium-loaded capillary, wherein said device provides at least parts per trillion (ppt) level detection sensitivities.